$$\text{Percent moisture} = \frac{(V_T - V_A) \times e \times 100}{W_s}$$

where:

e=Water equivalence of the Karl Fischer reagent determined as directed in paragraph (c)(1) of this section;

V_T=Milliliters of Karl Fischer reagent used;

V_A=Milliliters of Karl Fischer reagent equivalent to the 20 milliliters of solvent A, determined as directed in paragarph (c)(3) of this section;

W_s=Weight of the sample in milligrams.

(2) *Procedure 2.* Add an excess of Karl Fischer reagent to the sample, start the stirrer, and titrate to the endpoint with methanol solution. Calculate the percent moisture in the sample as follows:

(i) For powders:

$$\text{Percent moisture} = \frac{(V_T - V_m f) \times e \times 100}{W_s}$$

(ii) For oils and ointments:

$$\text{Percent moisture} = \frac{(V_T - V_m f - V_B) \times e \times 100}{W_s}$$

where:

V_T=Milliliters of Karl Fischer reagent used;

V_m=Milliliters of methanol solution used;

f=Milliliters of Karl Fischer reagent equivalent to each milliliter of methanol solution determined as directed in paragraph (c)(2) of this section.

V_B=Milliliters of Karl Fischer reagent equivalent to the 10 milliliters of solvent B determined as directed in paragraph (c)(3) of this section;

e=Water equivalence of the Karl Fischer reagent determined as directed in paragraph (c)(1) of this section;

W_s=Weight of the sample in milligrams.

(3) *Procedure 3.* Add about 20 milliliters of solvent A to a dry titrating vessel and proceed as directed in titration procedure 1 or 2. Disregard the volume of reagents used to determine the endpoint. Promptly introduce an accurately weighed or measured quantity of sample into the titrating vessel and titrate to the endpoint using either titration procedure 1 or 2 without additional solvents. Calculate the percent moisture in the sample as follows:

(i) If titration procedure 1 is used:

$$\text{Percent moisture in weighed samples} = \frac{V_T \times e \times 100}{W_s}$$

$$\text{Percent moisture in aerosols} = \frac{V_T \times e}{\text{Milliliters of sample} \times 10}$$

Percent moisture in hygroscopic powders=

$$\text{Percent moisture in hygroscopic powders} = \frac{(V_T - V_b) \times e \times 100}{W_s}$$

(ii) If titration procedure 2 is used:

$$\text{Percent moisture in weighed samples} = \frac{(V_T - V_m f) \times e \times 100}{W_s}$$

$$\text{Percent moisture in aerosols} = \frac{(V_T - V_m f) \times e}{\text{Milliliters of sample} \times 10}$$

$$\text{Percent moisture in hygroscopic powders} = \frac{(V_T - V_m f - V_b) \times e \times 100}{W_s}$$

where:

V_T=Milliliters of Karl Fischer reagent used;

V_m=Milliliters of methanol solution used;

f=Milliliters of Karl Fischer reagent equivalent to each milliliter of methanol solution determined as directed in paragraph (c)(2) of this section;

V_b=Karl Fischer equivalent (in milliliters) of the methanol used as a sample solvent;

e=Water equivalence of the Karl Fischer reagent determined as directed in paragraph (c)(1) of this section.

[39 FR 18944, May 30, 1974, as amended at 48 FR 51292, Nov. 8, 1983; 50 FR 41679, Oct. 15, 1985; 51 FR 22071, June 18, 1986; 51 FR 27532, Aug. 1, 1986]

§ 436.202 pH.

(a) *Apparatus.* A suitable potentiometer fitted with two electrodes, one being constructed of glass and sensitive to hydrogen ion activity and the other being a calomel or a silver/silver chloride reference electrode. A combination electrode with glass electrode and reference electrode contained in the same system may be used.

(b) *Standardization.* Select two standard buffer solutions such that the expected pH value of the sample is within their pH range and is also within 2 pH

unit of one of the standard buffer solutions. Standardize the pH meter with the two buffer solutions. Make any necessary adjustment of the meter if the observed pH value of either standard solution differs by more than 0.05 pH units of its known value.

(c) *Sample preparation.* If necessary, dilute the sample with carbon dioxide-free distilled water to the concentration specified in the individual section for each antibiotic.

(d) *Test procedure.* Determine the pH of the sample at 25°±2° C. Rinse the electrode(s) between determinations first with distilled water and then with a portion of the next sample to be tested. Store electrode(s) with tips immersed in water.

[39 FR 18944, May 30, 1974, as amended at 42 FR 29857, June 10, 1977; 42 FR 31449, June 21, 1977]

§ 436.203 Crystallinity.

Use the method specified in the individual section for each antibiotic.

(a) *Method 1.* To prepare the sample for examination, mount a few particles in mineral oil on a clean glass slide. Examine the sample by means of a polarizing microscope. The particles reveal the phenomena of birefringence and extinction positions on revolving the microscope stage.

(b) *Method 2.* Proceed as directed in paragraph (a) of this section, except to prepare the sample for examination, mount a few particles in mineral oil, add 1 drop of ethyl alcohol, and allow to react for about 30 seconds.

§ 436.204 Iodometric assay.

(a) *Reagents.* (1) 0.01*N* Sodium thiosulfate (2.482 grams $Na_2S_2O_35H_2O$ and 125 milligrams Na_2CO_3 per liter).

(2) 1.0*N* Sodium hydroxide.

(3) 1.2*N* Hydrochloric acid.

(4) 0.01*N* Iodine solution (prepared from 0.1*N* iodine U.S.P.).

(5) Starch iodide paste, T.S. (U.S.P.).

(b) *Preparation of sample and working standard solutions*—(1) *Workingstandard solutions.* From the following table, select the initial solvent, diluent, and final concentration as listed for each antibiotic working standard. Dissolve and dilute an accurately weighed portion to the specified final concentration and proceed as directed in paragraphs (c) and (d) of this section.

Antibiotic	Initial solvent	Diluent (solution number as listed in § 436.101(a))	Final concentration in units or milligrams of activity per milliliter of standard solution
Amoxicillin	None	Distilled water	1.0 mg.
Ampicillin	do	do	1.25 mg.
Cephalexin	do	do	2 mg.
Cephaloridine	do	do	2 mg.
Cephalothin	do	do	2 mg.
Cephapirin	do	do	2 mg.
Cloxacillin	do	do	1.25 mg.
Cyclacillin	do	do	1.0 mg.
Dicloxacillin	do	1	1.25 mg.
Methicillin	do	1	1.25 mg.
Nafcillin	do	1	1.25 mg.
Oxacillin	do	1	1.25 mg.
Penicillin G	do	1	2,000 units.
Penicillin V potassium	do	1	2,000 units.

(2) *Bulk antibiotic solutions.* From the following table, select the initial solvent, diluent, and final concentration as listed for each antibiotic. Dissolve an accurately weighed aliquot (approximately 30 to 60 milligrams) of the sample, dilute to the appropriate final concentration, and proceed as directed in paragraphs (c) and (d) of this section.

Antibiotic	Initial solvent	Diluent (solution number as listed in § 436.101(a))	Final concentration in units or milligrams of activity per milliliter of sample
Amoxicillin trihydrate	None	Distilled water	1.0 mg.
Ampicillin	do	do	1.25 mg.
Ampicillin sodium	do	1	1.25 mg.
Ampicillin trihydrate	do	Distilled water	125 mg.